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Investigation of the use of luminescent markers as gunshot residue indicators

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Abstract: The addition of luminescent markers into ammunition cartridges is an appealing proposal to achieve a greater individual identification of the ammunition. The lanthanide elements present in these luminescent materials act as characteristic chemical markers that, also, offer the great benefit of making gunshot residues (GSR) collection easier. Although the use of luminescent markers offers numerous advantages, and can be a reliable future option in the cartridge ammunition market, little or nothing is known about the interaction of the marker with the ammunition compounds after the shot. For the forensic laboratories, it is of utmost importance to anticipate the commercialization of tagged luminescent ammunition by knowing the type of particles produced after the discharge. That is, to investigate if markers merge with GSR to form single particles or, on the contrary, they form individual particles that travel separately from the GSR particles. In this work, conventional ammunition cartridges tagged with two types of luminescent markers were shot. Then, the particles produced were visualized under UV light on the floor, clothing targets and the shooter. The luminescent particles spread over the floor determined the shooter position and the bullet trajectory. The illumination of the shot targets allows the visualization of the GSR patterns only using a portable UV lamp, avoiding the use of colorimetric test. The GSR particles were collected and subjected to SEM-EDX and Raman spectroscopy analysis to get information about their inorganic and organic composition. The results indicated that part of the marker and the GSR merge and travel together. With this, particles composed by the marker and

propellant organic compounds can be identified by Raman spectroscopy, and the unequivocal identification of the GSR and the marker can be achieved by SEM-EDX. Consequently, the luminescent particles detected under UV light improve the evidence collection and offer information about the GSR and the marker. Additionally, this study support the use of luminescent ammunition to study, for example, the transfer or persistence of GSR.

Keywords: Gunshot residues; Raman; SEM-EDX; Luminescent markers.

Graphical abstract:



1. Introduction

The products of the primer detonation and gunpowder combustion, as well as partially unburned gunpowder and metallic particles taken from the gun barrel, projectile and cartridge, comprise the so-called gunshot residues (GSR) [1], [2], [3], [4]. These residues spread out after leaving the gun and deposited on any surface in the shooting surroundings. Therefore, GSR are commonly used to reconstruct a firearm-related offense, helping to confirm (or deny) the implication of a suspect, a suicide hypothesis, to estimate firing distance, to identify bullet holes, among others. For these reasons, GSR are physical evidence with great forensic value [1], [2], [3], [4], [5].

Nowadays, the recommended technique for GSR characterization is Scanning Electron Microscopy with Energy Dispersive X-ray spectroscopy (SEM-EDX) [6]. The GSR particles from conventional ammunition are identified by SEM-EDX by their spheroidal shape and the simultaneous presence of Sb, Ba and Pb at the same particle [4]. However, the introduction in the market of lead-free ammunition has resulted in the development of new analytical methods focused on the additional identification of the organic compounds of GSR. For this, Raman spectroscopy, a non-destructive spectroscopic technique that offers information about both organic and inorganic samples, was proposed to identify the organic components present in GSR particles. In turn, López-López et al. [7] showed that the Raman spectra of the macroscopic GSR particles have high similarity to the unburned gunpowder and, therefore, may help to trace ammunition. Furthermore, an GSR-discrimination based on the stabilizers used in the propellant (diphenylamine or ethylcentralite) proved to be efficient. In other work, the same research group [8] showed that the weapon memory effect is insignificant when analyzing the organic part of the GSR particles by Raman, reducing the risk of contamination by previous shots. Meanwhile, Abrego et al. [9] used an adhesive polytetrafluoroethylene tape lift for collecting GSR particles from shooters' hands to analyze the particles by a confocal Raman microscope. In this study, the gunpowder stabilizers were also detected in the GSR particles collected. Bueno et al. [10] used a near-infrared confocal Raman microscope and chemometrics to discriminate GSR particles from two different ammunition calibers (9 mm and 0.38). More recently, Raman imaging was used to characterize the organic components of the GSR particles [11], [12]. In short, Raman spectroscopy, stand out since it has numerous advantages over other techniques. As example, it does not require sample preparation (the measurements can be performed directly on SEM stubs commonly used to collect the

GSR particles as well as on a blood-stained victim's clothes) [12], performs nondestructive analysis (important to characterize forensic evidence) and provides spectra that can be compared with the profile of known samples in a fast way. Additionally, the mapping of the particle can be performed with some imaging equipment, allowing to visualize the spectra of the particle at different points of the sample [12].

When it comes to GSR analysis, a source of false negatives that should be considered is the possibility of non-collecting the GSR. Due the micron size of these particles, many times the forensic expert collect the sample without visualizing the residues. In this context, Weber et al. [13], [14], [15], [16], [17], [18], [19] proposed the addition of luminescent markers into ammunition cartridges to assist the visual detection of their residues, their collection and the crime scene investigation. Luminescent markers, when added into ammunition, allow the visual detection of luminescent residues directly in loco, using only a portable UV lamp. Furthermore, once the lanthanide elements present in these materials are uncommon in the environment, they act as chemical markers and can assist in the identification of GSR particles, especially those coming from lead-free ammunition [16], [17], [18], [19]. Besides, different emission colors can be used to differentiate ammunition by usage type (e.g. civilian, police or military) [18].

Although the characterization of isolated luminescent markers (after shots with conventional [14], [15] and NTA [14], [16], [17], [18], [19]) is well established by using techniques such as SEM-EDX and emission spectroscopy, the interaction of these markers with the ammunition compounds after the shot was not studied to date. Then, it is of utmost importance to know if the dispersion of the luminescent markers is similar to the GSR particles by knowing if they give rise to two different types of particles (marker and GSR particles), or, on the contrary, they lead to particles consisting of a mixture of GSR and lanthanides. In other words, it is important to assess whether the markers improve the collection of organic and inorganic GSR particles or not.

This work aimed to study the GSR particles produced after the discharge of conventional ammunition cartridges tagged with two types of luminescent markers. Firstly, the visualization of the luminescent particles under UV light on the floor, clothing targets and shooters was pursued. Then, the particles were collected using SEM stubs and characterized by the reference technique, SEM-EDX, and also, by Raman spectroscopy, which is one of the most promising techniques for the analysis of the organic components of the GSR.

2. Material and methods

2.1. Sample preparation

The bullets of 9 × 19 mm semi-jacketed hollow point conventional ammunition manufactured by Sellier&Bellot (Czech Republic) were removed from the cartridges using a bullet puller. Then, a green ($[Y_{0,85}Yb_{0,10}Tb_{0,05}(BTC)]$) or red ($[Y_{0,95}Eu_{0,05}(BTC)]$) luminescent marker (wherein BTC = trimesic acid), prepared as previously reported in Ref. [19], was added to the gunpowder (5 wt%). The handloading process of the tagged ammunition was performed in a press.

The floor of a shooting range was covered with a 10-m black paper strip in order to keep the floor surface where the GSR particles are going to be collected as clean as possible. Then, a shot was performed using a Glock G17 pistol and an ammunition cartridge tagged with the red marker. After the shot, the shooter and the floor were observed under UV radiation ($\lambda = 254$ nm) with a portable lamp (UVGL-58 Handheld UV Lamp, UVP). Particles were collected from the shooter's hands, firearm and the floor (around 0.1, 0.3, 1.2, 2 and 8 m from the shooting point) with 13 mm diameter aluminum SEM stubs with adhesive carbon tape (one stub for the hands, one stub for the firearm, and one stub for each distance). Then, the black paper strip was replaced by a new one and the same procedure was repeated for the ammunition tagged with the green marker (different shooter and G17 pistol). In both cases, the pistols were not previously cleaned to provide a more realistic scenario. The stubs ($n = 14$) used to collect the particles at the different distances, the shooter's hands and the firearms were then analyzed by SEM-EDX.

In a second step, 35 × 35 cm cardboard targets covered with black fabric were shot at five different distances (0.1, 0.3, 1.2, 2 and 8 m) with the two types of tagged ammunition cartridges. Photos (Fuji Finepix S100 fs) of the targets were taken under UV radiation ($\lambda = 254$ nm) to visualize the GSR patterns. The GSR particles on the targets were collected with SEM stubs (one stub for each target), and then, analyzed using Raman spectroscopy. The particles that emit luminescence were focused under the Raman microscope, and analyzed.

2.2. Instrumentation

The scanning electron microscope Quanta 200 from FEI (Brno, Czech Republic) with an energy dispersive X-ray spectrometer EDAX, (Si(Li) detector, SUTW—Super Ultra Thin Window, resolution 135 eV for Mn K α) was used at low-vacuum. The automatic identification of GSR was performed with a Gunshot program (GSR-XT V3.1 for FEI). A 160 \times magnification and an accelerating voltage of 25 kV were employed. The entire area of each stub was analyzed. The agreement of the chemical composition of a particle (resulting from its X-ray spectrum) with the class assigned to it by the program was checked and corrected manually.

A DXR Raman Microscope from Thermo Scientific (Waltham, MA) controlled by the Thermo Scientific Omnic for dispersive Raman 8.1 software was employed. A laser of 532 nm with potency ranging from 1 to 10 mW and a confocal pinhole size of 50 μ m was used. The microscope objective was set to 50 \times magnification, and at least 10 spectra were acquired for each particle. Spectra were acquired with fluorescence correction.

3. Results and discussion

3.1. Visualization of luminescent particles under UV radiation

After the shooting of the ammunition cartridge tagged with the red marker, a luminescent trail of particles of approximately 9 m was observed when the black paper laid on the floor was illuminated with the portable UV lamp ($\lambda = 254$ nm). The same result was observed for the ammunition cartridge tagged with the green marker. This fact is in agreement with the observations previously reported in Ref. [18] for tagged non-toxic ammunition. As expected, the amount of luminescent particles spread over the floor decreases as the distance from the shooter position increases. In fact, the higher concentration of these particles occurred at the first 30 cm from the shooter position and gradually reduced over the next 3.3 m. The visualization of this trail of particles on the floor resulted in the identification of the shooter position and the estimation of the bullet trajectory. This information could be of extraordinary assistance at the crime scene for crime reconstruction. Besides the floor, the luminescent particles were also visualized on other surfaces near the shooter position, including the shooter (hands, head, clothing,

shooter's shoes and belt, etc.) and the firearm. Additionally, the finding of the empty cartridge case or the bullet, both important ballistic evidence to collect at the crime scene, was facilitated due to their fluorescent nature. Fig. 1 depicts several luminescent particles on different surfaces illuminated with the UV lamp.

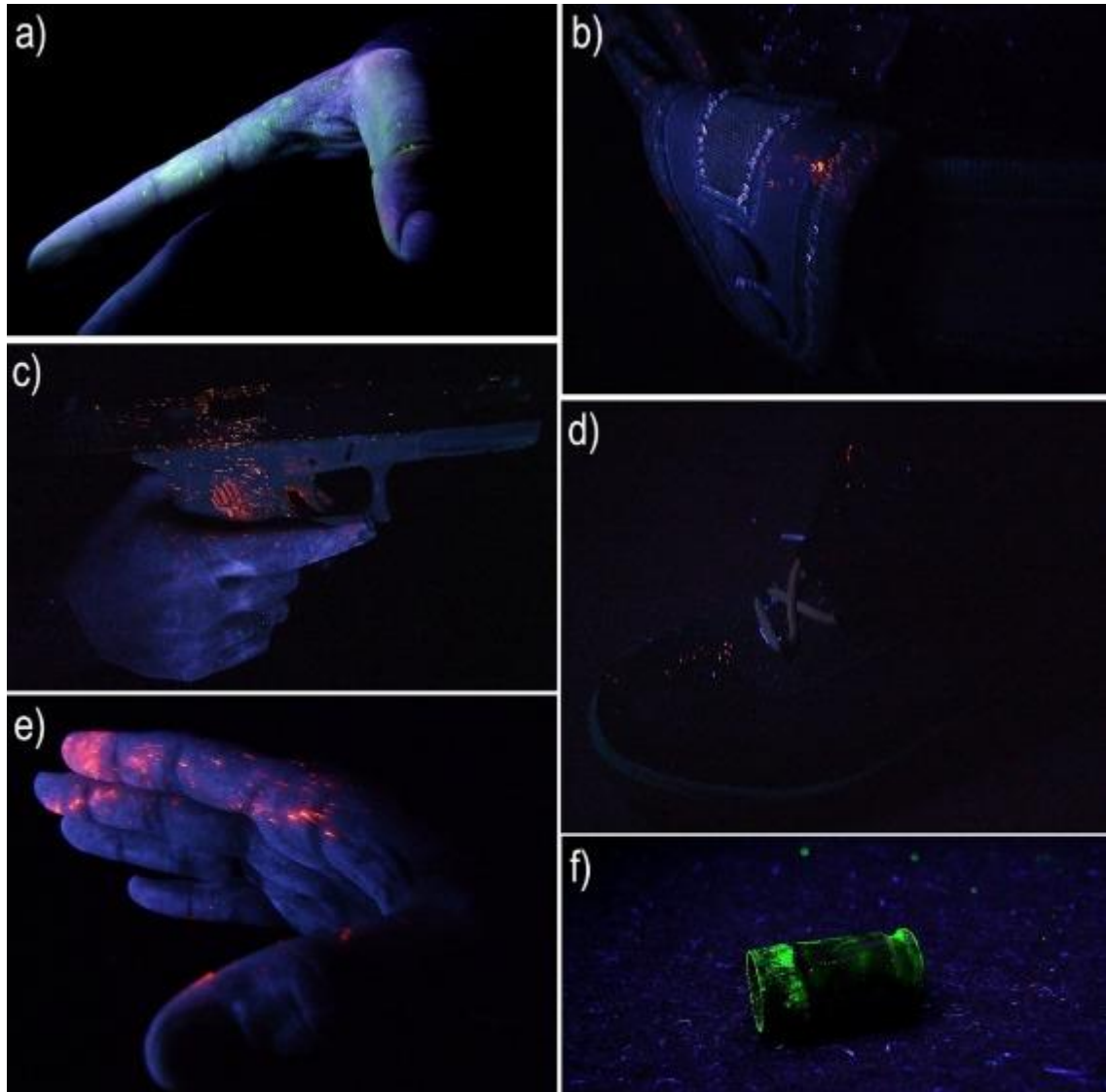


Fig. 1. Images under UV light ($\lambda = 254 \text{ nm}$) of luminescent red or green particles resulting after the shooting of tagged ammunition on (a) and (e) shooters' hand, (b) shooters' belt, (c) firearm, (d) shooter's shoe and (f) cartridge case.

Besides the transference of GSR particles to the shooter or to different objects at the scene, these particles can be transferred to the victim. The particles that reached the target (victim) creates a GSR pattern that depends on different factors (e.g. type of ammunition and firearm, firing angle, atmospheric conditions, etc.), being the firing

distance the most important determinant [18], [20]. The study of the GSR patterns on the victim's clothing is performed on the forensic laboratories to estimate the shooting distance (muzzle to target distance), which has an extensive forensic significance for the reconstruction of a firearm-related offense. However, the visualization of the GSR patterns is not always an easy task, and tests based on color chemical reactions (e.g., Walker test, Marshal and Tewari test, sodium rhodizonate test, etc.) are used to reveal the presence of nitro groups or heavy metals present in the GSR particles [21].

Therefore, after the visualization of the luminescent particles on the floor, five targets covered with black clothing were shot with the two tagged ammunitions (red and green markers) at different distances (0.1, 0.3, 1.2, 2 and 8 m) to evaluate if the GSR patterns can be revealed using only the UV lamp. Fig. 2 shows the targets shot with the two tagged ammunitions under UV illumination. For both tagged ammunitions, the targets shot at 10 cm showed a great amount of luminescent particles and concentric circles left by the plume of gases. As the firing distance increases, these circles disappeared and the amount of particles decreases, being also these particles more dispersed. At 30 cm the concentric circles are not visualized on the targets but there was also a great amount of particles. The luminescent patterns were easily visualized also on the targets shot at 1.2 m, despite the amount of particles declined considerably compared to the previous targets and, also, these particles were more dispersed. Still, a few particles were visualized on the targets shot at 2 m. However, without considering the dirt ring around the entry hole, only three small particles were found (two red particles and one green particle) on the targets shot at 8 m. In all cases, the presence of the bullet wipe residues allowed to visualize the entry holes, due to their luminescent nature.

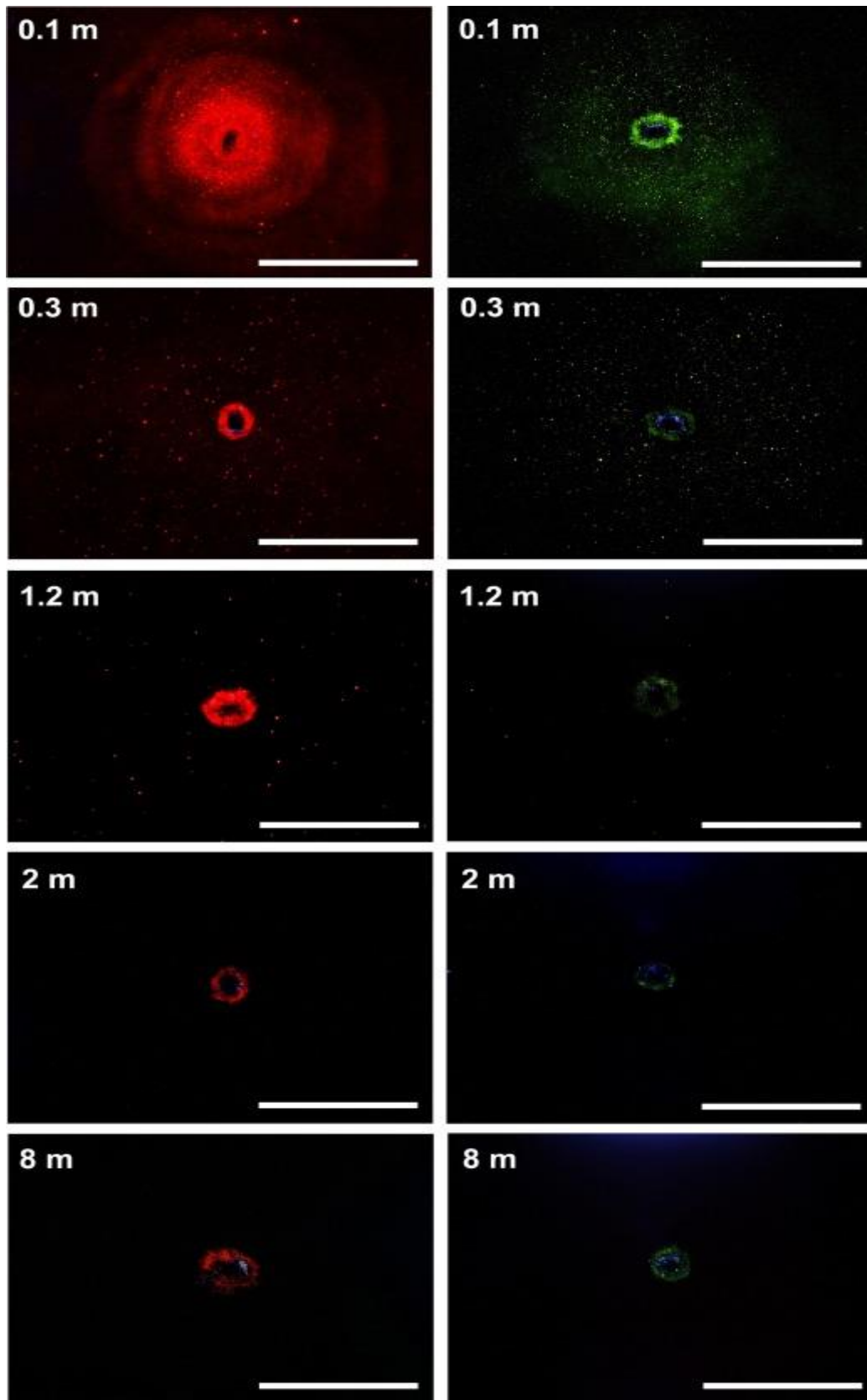


Fig. 2. Images of targets shot with 9 mm conventional ammunition tagged with red (left) and green (right) markers under UV radiation ($\lambda = 254$ nm). Muzzle-to-target distances were 0.1, 0.3, 1.2, 2 and 8 m (top to bottom). White bars represent 4 cm scales.

The results obtained indicate that, for tagged ammunition, it is possible to visualize GSR patterns using only a portable UV lamp. This methodology does not require the use of colorimetric tests, showing clear benefits in terms of time, cost and no exposure to chemicals. Additionally, no sample damage or manipulation is required. It is also important to mention that the GSR pattern would be directly visualized in situ, at the crime scene, on the victim's body, provided that low light conditions are fulfilled [18].

3.2. SEM-EDX and Raman analysis of luminescent particles

In order to evaluate if luminescent markers can act as a reliable probe to locate GSR particles, it is necessary to know if luminescent markers can be present with both inorganic and organic GSR compounds and, in that event, how far from the shooting point can these particles be found. For this, samples collected at different shooting distances were analyzed by SEM-EDX and Raman spectroscopy to investigate if markers merge with GSR to form single particles or, on the contrary, they form individual particles that travel separately from the GSR particles.

3.2.1. SEM-EDX analysis

The particles collected with the adhesive stubs on the shooters' hands, firearms and on the floor were analyzed by SEM-EDX. Fig. 3, Fig. 4 show the SEM-EDX results obtained for the particles collected after the shooting of the ammunition cartridge tagged with the red (95% Y and 5% Eu) and green (85% Y, 10% Yb and 5% Tb) markers, respectively. For best understanding, the particles analyzed were classified into five groups, according to the composition: (i) Type I: characteristic of GSR (PbBaSb particles); (ii) Type II: characteristic of GSR and marker (PbBaSbYEu particles for the red marker and PbBaSbYTbYb particles for the green marker); (iii) Type III: characteristic of GSR and indicative identification of the marker (PbBaSbY particles for the red marker and PbBaSbY particles for the green marker); (iv) Type IV: characteristic of the marker (YEu particles for the red marker and YTbYb particles for the green marker); (v) Type V: indicative of luminescent marker (Y particles for the red marker and Y or Tb particles for the green marker); (vi) others: particles not included in Types I–V.

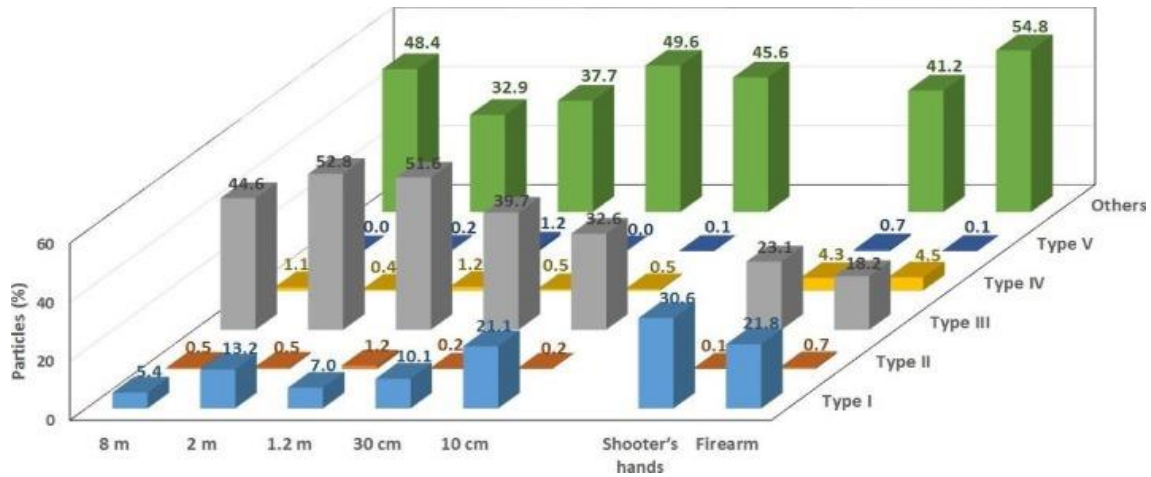


Fig. 3. SEM-EDX results obtained for the particles collected after the shootings performed with the conventional ammunition tagged with the red marker (95% Y, 5% Eu). Type I: characteristic of GSR (PbBaSb particles); Type II: characteristic of GSR and red marker (PbBaSbYEu particles); Type III: characteristic of GSR and indicative identification of red marker (PbBaSbY or PbBaSbEu particles); Type IV: characteristic of red marker (YEu particles); Type V: indicative of luminescent marker (Y or Eu particles); Type 0: particles not included in Types I–V. Total of particles analyzed: 8 m, 186 particles; 2 m, 547 particles; 1.2 m, 488 particles; 30 cm, 587 particles; 10 cm, 1016 particles; shooter's hands, 3512 particles; and firearm, 896 particles.

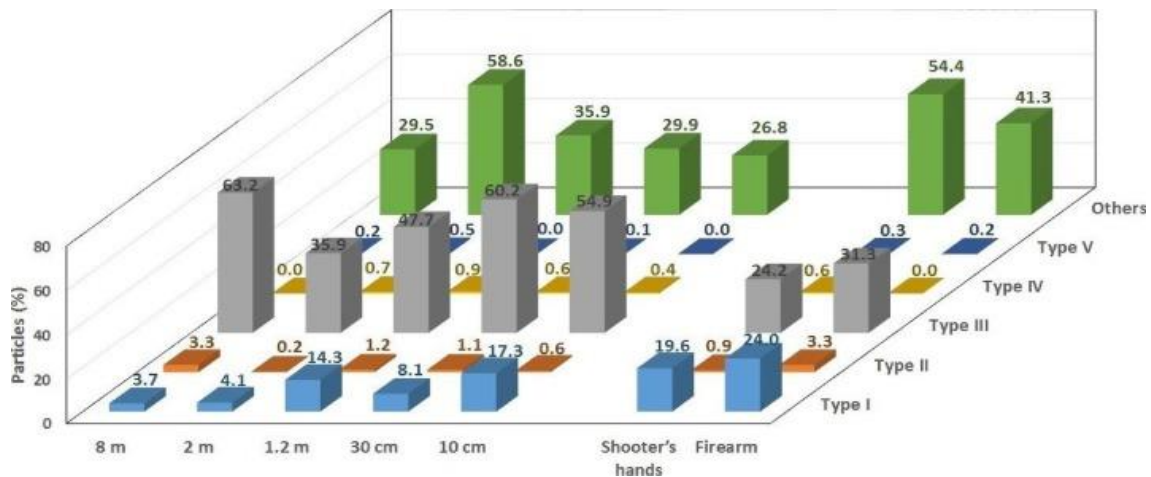


Fig. 4. SEM-EDX results obtained for the particles collected after the shootings performed with the conventional ammunition tagged with the green marker (85% Y, 10% Yb, 5% Tb). Type I: characteristic of GSR (PbBaSb particles); Type II: characteristic of GSR and green marker (PbBaSbYTbYb particles); Type III: characteristic of GSR and indicative identification of green marker (PbBaSbY or PbBaSbTb particles); Type IV: characteristic of green marker (YTbYb particles); Type V: indicative of luminescent marker (Y or Tb particles); Type 0: particles not included in Types I–V. Total of particles analyzed: 8 m, 484 particles; 2 m, 418 particles; 1.2 m, 1626 particles; 30 cm, 1598 particles; 10 cm, 956 particles; shooter's hands, 1409 particles; and firearm, 521 particles.

In both tagged ammunitions, as expected, the number of particles detected by SEM-EDX highly varied depending on the collection point. The total of particles analyzed for the ammunition tagged with the red and green marker were: 8 m distance, 186 and 484 particles; 2 m, 547 and 418 particles; 1.2 m, 488 and 1626 particles; 30 cm, 587 and 1598 particles; 10 cm, 1016 and 956 particles; shooter's hands, 3512 and 1409 particles; and firearm, 896 and 521 particles. Then, the greatest variation regarding the number of particles for the red ammunition was for the stub used to collect the particles at 8 m (186 particles) from the shooting point and the hand of the shooter (3512 particles). In the case of the green ammunition, the greatest variation observed regarding the number of particles was between the particles collected at 2 m (418 particles) and 1.2 m (1626 particles) from the shooter position. From all the particles detected at each stub, 45–67% were included into one of the five groups above mentioned for the particles coming from the ammunition cartridge tagged with the red marker; while 41–73% were included into these five types from the particles coming from the ammunition cartridge tagged with the green marker. The remained particles corresponded to compositions that were not considered in the study due to they do not unequivocally identify the GSR or do not offer information about the marker (Pb, Ba, Sb, Pb–Ba, Pb–Sb, Ba–Al, Si–Ca–Ba, Cu–Zn, Fe, etc.). The variation observed on the total number of particles depended on the number of particles present on the stubs that, apart from the logic dependence on the point of collection of the sample, is related to other parameters such as adhesive saturation or the dirt from the surface.

The results obtained for the particles coming from the ammunition tagged with the red marker and collected on the floor (Fig. 3) showed a percentage of PbBaSb particles (Type I) of 5–21%, while the percentage of particles that allow the identification of GSR and offer an indicative identification of the marker (PbBaSbY particles for the red marker, Type III) was 33–53%. This indicates that more PbBaSb particles combined with Y (element from the luminescent marker) were found. No particles with PbBaSbEu composition were detected, probably due to the major proportion of the Y in the marker. Particles that allowed the identification of the GSR and the marker (PbBaSbYEu, Type II particles) were detected in all cases, but in a low percentage (0.2–1.2%, which corresponds to 1–6 particles). Regarding to particles only composed by the marker (Type IV), they corresponded to 0.4–1.2% (2–6 particles) of the total of the particles detected. In the case of particles only composed by one of the elements of the marker (Y particles, Type V), they were only found at three distances (10 cm, 1 particle; 1.2 m, 6 particles; and 2 m, 1 particle); not having found Eu particles. These findings indicate that the

lanthanides of the marker tend to join particles coming from the ammunition in the detonation of the cartridge.

In the case of the samples collected on the shooter's hands and the firearm after the discharge of the ammunition tagged with the red marker (Fig. 3), an increase of Type I particles was observed (31% in hands and 22% in the firearm) compared with the results obtained for the particles collected at the floor. This result may be because the firearm was not cleaned before the shootings. Therefore, this greater percentage may be due to the accumulation of PbBaSb particles in the firearm from previous shootings with conventional ammunition cartridges. This also influences the number of particles found on the hand, due to its direct contact with the firearm. It is also very significant the increase of particles of marker (Type IV) in both cases (~4.5% in both cases), compared with the results obtained for the samples collected on the floor. This difference may be due to an inefficient crimp of the bullet with the cartridge (the crimp was manual). This could lead to a greater marker exhaust through the ejection port of the pistol.

The SEM-EDX results of the ammunition cartridge tagged with the green marker are summarized in Fig. 4. The results obtained were similar to those obtained with the red marker. For the samples collected on the floor, the percentages obtained were: 4–18% Type I particles (PbBaSb), 0.2–3.3% Type II particles (PbBaSbYTbYb), 36–63% Type III particles (PbBaSbY, no PbBaSbTb particles were found), <1% of Type IV particles (YTbYb, these particles were not found at 8 m), and <1% of Type V particles (1 particle of Y was detected at 30 cm, 2 m, and 8 m and 1 particle of Tb at 2 m). As above, there is a predominance of PbBaSb particles combined with Y over PbBaSb particles. However, for the ammunition tagged with the green marker, the particles that identify both GSR and the marker (Type II) considerably increased in all cases, compared with the results obtained with the red marker. As an example, the sample collected at 30 cm presented 18 particles of this type while, at the same distance, only 1 particle of this type was detected on the sample collected for the ammunition doped with the red marker. The exception to this trend was the sample collected at 2 m, where only one Type II particle was detected.

The samples collected on the shooter's hands and the firearm after the discharge of the ammunition tagged with the green marker also showed similar results to those obtained for the red marker. As can be noted in Fig. 4, there is a predominance on the Type III particles (24% and 31% for the hands and the firearm, respectively) over the particles characteristic of GSR (Type I, 19% and 24% for the hands and the firearm, respectively). The major difference between these samples and the ones obtained with

the red marker was observed on the particles that allow the characterization of GSR and the marker (Type II) and the particles only composed by the marker (Type IV). As in the samples collected on the floor, a modest increment of Type II particles was observed compared with the results obtained for the ammunition tagged with the red marker. In the case of the findings obtained for the Type II particles, they are in good agreement with the results obtained for the samples collected at the floor. This reinforces the hypothesis of a not efficient crimping of the ammunition tagged with the red marker.

Considering the results obtained for both tagged ammunitions, as expected, the percentage of GSR particles (Type I) decreased with the distance of collection. For example, from 10 cm to 8 m, the percentage of PbBaSb particles decreases from 21% (213 of 1016 particles analyzed) to 5% (9 of 186 particles analyzed) for the red marker and from 17% (162 of 959 particles analyzed) to 4% (19 of 484 particles analyzed) for the green marker. However, the tendency of the marker and the GSR particles to merge after shot, allied to the luminescent properties of the marker, improved the collection of luminescent GSR particles. With this, the percentage of particles Types II and III did not decrease, or even increased, with the distance. These results suggest that the markers act as luminescent probes for detecting GSR particles. This feature brings clear benefits for investigation of firearm-related crimes. For example, it is easier to find luminescent GSR particles away from the shooting point or even at unconventional locations performing a guided collection than a blind one.

3.2.2. Raman results

An exploratory analysis of the luminescent particles on the targets was performed by Raman spectroscopy to study if some of them could offer information about the organic part of the GSR and the marker. For this, SEM stubs were used to collect the luminescent particles and then, these particles were focused under the Raman microscope (the analysis of the stub stopped when three particles with GSR and marker Raman bands were found). Additionally, as-prepared markers and particles of unfired gunpowder coming from the ammunition were analyzed. Fig. 5, Fig. 6 depict the Raman spectra obtained for the as-prepared markers (red spectrum for the red marker and green spectrum for the green marker), the gunpowder without the markers (blue spectra), and the spectra of the particles analyzed at different targets (black spectra).

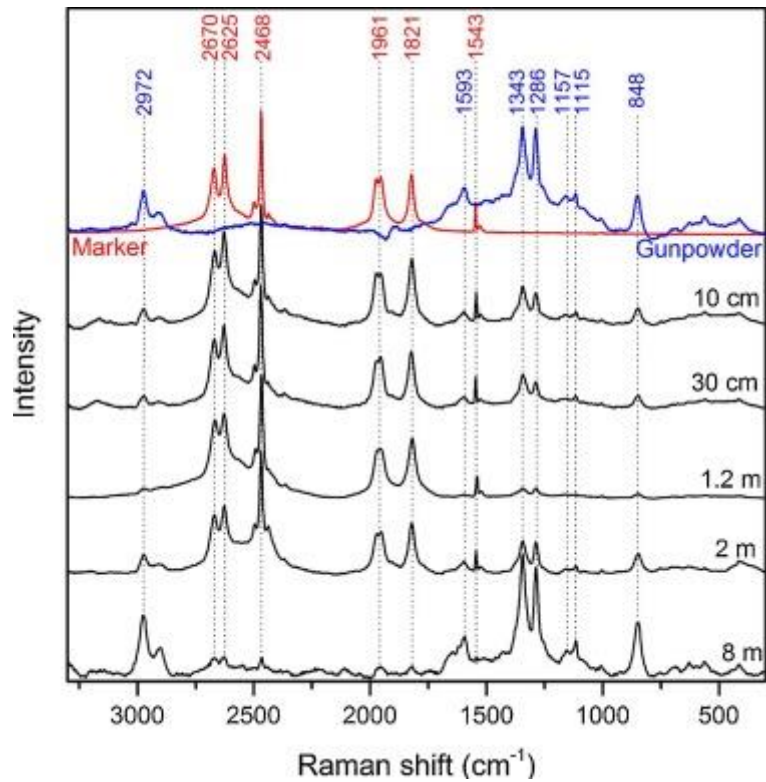


Fig. 5. Raman spectra of red marker (red line), unfired gunpowder from 9 mm conventional ammunition without marker (blue line) and luminescent particles from conventional ammunition tagged with the red marker (black lines). Raman conditions: laser at 532 nm, 50× magnification objective lens, confocal pinhole size of 50 μm . Spectral acquisition times and power: 0.5 s \times 3 acquisition, 2 mW (red marker); 10 s \times 5 acquisition, 8 mW (gunpowder and luminescent particles on targets).

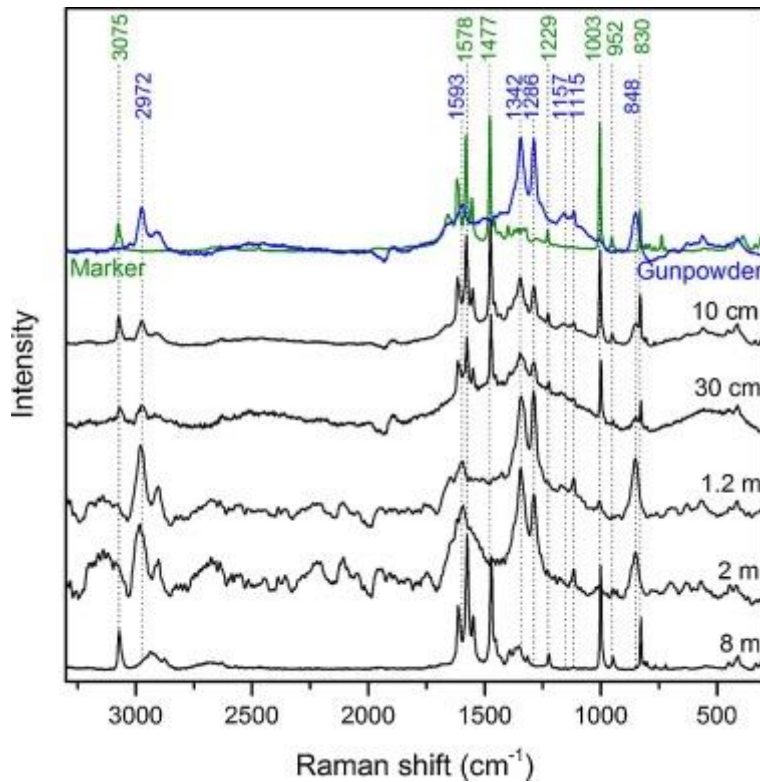


Fig. 6. Raman spectra of green marker (green line), unfired gunpowder from 9 mm conventional ammunition without marker (blue line) and luminescent particles from conventional ammunition tagged with the green marker (black lines). Raman conditions: laser at 532 nm, 50× magnification objective lens, confocal pinhole size of 50 μm . Spectral acquisition times and power: 10 s \times 20 acquisition, 3 mW (green marker); 10 s \times 5 acquisition, 8 mW (gunpowder and luminescent particles on targets).

The targets shot with the ammunition cartridge tagged with the red marker (Fig. 5) presented mostly particles with bands coming from the luminescent marker and the gunpowder. However, several particles that only show bands of one or other compound were also detected. The first type of particles, those that showed bands of luminescent marker and the gunpowder, were identified at the targets shot at 0.1, 0.3, 1.2 and 2 m. These particles presented bands at around 2670, 2625, 2468, 1961, 1821 and 1543 cm^{-1} coming from the red marker; and bands from nitrocellulose (the main constituent, and active component, of smokeless gunpowders) at around 2972, 1286, 1157, 1115 and 848 cm^{-1} . Furthermore, N-nitrosodiphenylamine (1593 cm^{-1}) and 2-nitro-diphenylamine (1343 cm^{-1}) were observed [7]. As expected, with increasing the shooting distance, it was more difficult to find particles containing both components (marker and gunpowder). As example, just a small particle ($\sim 160 \mu\text{m}$ diameter) that presented both luminescent marker and gunpowder bands was found on the target shot at 8 m and, as can be seen,

the signal coming from the marker is quite weak. This fact may be due to a small amount of marker present in the particle or because the marker was in a position of the particle where the laser cannot be properly focused. However, due the small number of particles found, it was difficult to eliminate the possibility of contamination from other shots or even by handling the targets.

Regarding the results obtained with the green marker (Fig. 6), particles with bands coming from the marker (3075, 1578, 1477, 1229, 1003, 952 and 830 cm^{-1}) and the gunpowder (2972, 1593, 1342, 1286, 1157, 1115 and 848 cm^{-1}) were also detected at the targets shot at 10 and 30 cm. In the case of the targets shot at 1.2 and 2 m, although luminescent particles have been visualized under UV radiation, the Raman spectra of these particles presented only bands from the gunpowder. As in the previous case of the target shot at 8 m with the ammunition cartridge tagged with red marker, possible explanations of this fact may be that the luminescent marker were in an internal layer or in a position where the laser cannot be focused (e.g. superficial plane over the lateral face). For the target shot at 8 m, just a particle that presented marker bands was found.

Thus, Raman and SEM-EDX results suggest that, after the shooting, a fraction of the marker joins the GSR. With this, an efficient collection of GSR is ensured collecting luminescent particles. Meanwhile, another fraction of residues (marker and GSR) remains as isolated particles. However, some of these particles experience similar trajectories. In this case, performing the collection at places where luminescent particles are visualized also increases the chances of collecting GSR. In both cases, luminescent marker can improve the quality of the collection. In that sense, one very valuable practical utility of luminescent ammunition could be to use them for the study of the transfer and persistence of GSR.

4. Conclusions

9 mm conventional ammunition cartridges tagged with two different luminescent markers were fired to investigate the composition of the particles produced after the discharge. The luminescent particles spread over the floor resulting in a luminescent trail of particles of approximately 9 m that helps to estimate the shooter's position and bullet trajectory. The higher concentration of these particles occurred at the first 30 cm from the shooter position and was gradually reduced over the next 3.3 m. Additionally, the

illumination of the clothing targets shot indicate the possibility to visualize GSR patterns only using a portable UV lamp, avoiding the use of colorimetric tests.

The SEM-EDX analysis of the residues collected on the floor showed a majority of particles with a composition that identified the GSR while offer an indicative identification of the marker (Type III particles). Moreover, the percentage of these particles at the five distances studied were similar, probably because the collection of particles was performed where luminescent particles were visualized. Additionally, some particles of Type II (characteristic of GSR and marker) were found at different distances. The samples collected on the shooter hands and the firearm presented a higher percentage of PbBaSb particles, probably due to a contamination of the firearms which were not cleaned before the shootings.

The analysis of the luminescent particles by Raman spectroscopy indicated that particles composed by the marker and gunpowder can be found at least until 8 m, in the case of the ammunition tagged with the red marker, and until 2 m for the ammunition tagged with the green marker. This result strongly suggests that markers tend to merge to GSR. Additionally, the visualization of the luminescent particles under UV light, in fact, improves the sample collection and, therefore, increases the possibility of finding GSR particles.

The combination of Raman and SEM-EDX on the analysis of residues coming from luminescent tagged ammunition provided information about the organic and inorganic components of both GSR and luminescent marker. Although it is not possible to rule out conclusively a possible contamination effect, due to the performance of the experiments in a gallery where there is an obvious presence of particles PbBaSb, it is clear that this possible interference should not be considered. Otherwise, the results obtained at the second experiment (ammunition tagged with green marker) should show a significant increase of PbBaSb particles compared with the first experiment (ammunition tagged with red marker); and this did not happen.

Finally, it should be mentioned that the results obtained in this article depend on the analyst skills (visualization of luminescent particles before collection), the adherent capacity of the stubs as well as the possible dirt on the collection surface. Also, the fact of using tagged hand loaded ammunition is undoubtedly an effect to consider. Therefore, the distances reached by the particles, as well as the number of particles found at each distance, should be considered approximate.

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References

- [1] O. Dalby, D. Butler, J.W. Birkett, Analysis of gunshot residue and associated materials—a review, *J. Forensic Sci.* 55 (2010) 924–943, doi:<http://dx.doi.org/10.1111/j.1556-4029.2010.01370.x>.
- [2] L. Garofano, M. Capra, F. Ferrari, G.P. Bizzaro, D. Di Tullio, M. Dell’Olio, A. Ghitti, Gunshot residue: further studies on particles of environmental and occupational origin, *Forensic Sci. Int.* 103 (1999) 1–21, doi:[http://dx.doi.org/10.1016/S0379-0738\(99\)00035-3](http://dx.doi.org/10.1016/S0379-0738(99)00035-3).
- [3] F. Saverio Romolo, P. Margot, Identification of gunshot residue: a critical review, *Forensic Sci. Int.* 119 (2001) 195–211, doi:[http://dx.doi.org/10.1016/S0379-0738\(00\)00428-X](http://dx.doi.org/10.1016/S0379-0738(00)00428-X).
- [4] A.J. Schwoeble, D.L. Exline, *Current Methods in Forensic Gunshot Residue Analysis*, CRC Press LLC, Boca Raton, FL, 2000.
- [5] S. Basu, Formation of gunshot residues, *J. Forensic Sci.* 27 (1982) 11453, doi:<http://dx.doi.org/10.1520/JFS11453J>.
- [6] ASTM (American Society for Testing and Materials), *Standard Guide for Gunshot Residue Analysis by Scanning Electron Microscopy/Energy Dispersive X-ray Spectrometry (ASTM E 1588-08)*, ASTM, 2008, doi:<http://dx.doi.org/10.1520/E1588-10E01>.
- [7] M. López-López, J.J. Delgado, C. García-Ruiz, Ammunition identification by means of the organic analysis of gunshot residues using Raman spectroscopy, *Anal. Chem.* 84 (2012) 3581–3585, doi:<http://dx.doi.org/10.1021/ac203237w>.

- [8] M. López-López, J.J. Delgado, C. García-Ruiz, Analysis of macroscopic gunshot residues by Raman spectroscopy to assess the weapon memory effect, *Forensic Sci. Int.* 231 (2013) 1–5, doi:<http://dx.doi.org/10.1016/j.forsciint.2013.03.049>.
- [9] Z. Abrego, N. Grijalba, N. Unceta, M. Maguregui, A. Sanchez, A. Fernández-Isla, M.A. Goicolea, R.J. Barrio, A novel method for the identification of inorganic and organic gunshot residue particles of lead-free ammunitions from the hands of shooters using scanning laser ablation-ICPMS and Raman microspectroscopy, *Analyst* 139 (2014) 6232–6241, doi:<http://dx.doi.org/10.1039/C4AN01051E>.
- [10] J. Bueno, V. Sikirzhytski, I.K. Lednev, Raman spectroscopic analysis of gunshot residue offering great potential for caliber differentiation, *Anal. Chem.* 84 (2012) 4334–4339, doi:<http://dx.doi.org/10.1021/ac203429x>.
- [11] J. Bueno, I.K. Lednev, Raman microspectroscopic chemical mapping and chemometric classification for the identification of gunshot residue on adhesive tape, *Anal. Bioanal. Chem.* 406 (2014) 4595–4599, doi:<http://dx.doi.org/10.1007/s00216-014-7874-9>.
- [12] M. López-López, M.Á.F. de la Ossa, C. García-Ruiz, Fast analysis of complete macroscopic gunshot residues on substrates using Raman imaging, *Appl. Spectrosc.* 69 (2015) 889–893, doi:<http://dx.doi.org/10.1366/14-07816>.
- [13] I.T. Weber, A.J.G. Melo, M.A.M. Lucena, M.O. Rodrigues, S. Alves Júnior, Processo de Obtenção de Munição Luminescente e Processo de Detecção de Resíduos de Tiro – BR Patent WO/2010/105326, 2010.
- [14] I.T. Weber, A.J.G. Melo, M.A.M. Lucena, M.O. Rodrigues, S. Alves Júnior, High photoluminescent metal-organic frameworks as optical markers for the identification of gunshot residues, *Anal. Chem.* 83 (2011) 4720–4723, doi:<http://dx.doi.org/10.1021/ac200680a>.
- [15] I.T. Weber, I.A.A. Terra, A.J.G. Melo, M.A.M. Lucena, K.A. Wanderley, C.O. Paiva-Santos, S.G. Antônio, L.A.O. Nunes, F.A.A. Paz, G.F. de Sá, S. Alves Júnior, M.O. Rodrigues, Up-conversion properties of lanthanide-organic frameworks and how to track ammunitions using these materials, *RSC Adv.* 2 (2012) 3083, doi:<http://dx.doi.org/10.1039/c2ra01214f>.
- [16] M.A.M. Lucena, G.F. de Sá, M.O. Rodrigues, S. Alves Júnior, M. Talhavini, I.T. Weber, ZnAl₂O₄-based luminescent marker for gunshot residue identification and

ammunition traceability, *Anal. Methods* 5 (2013) 705, doi:<http://dx.doi.org/10.1039/c2ay25535a>.

[17] M.A.M. Lucena, M.O. Rodrigues, C.C. Gatto, M. Talhavini, A.O. Maldaner, S. Alves Júnior, I.T. Weber, Synthesis of [Dy(DPA)(HDP)] and its potential as gunshot residue marker, *J. Lumin.* 170 (2016) 697–700, doi:<http://dx.doi.org/10.1016/j.jlumin.2015.04.010>.

[18] I.T. Weber, A.J.G. Melo, M.A.M. Lucena, E.F. Consoli, M.O. Rodrigues, G.F. de Sá, A.O. Maldaner, M. Talhavini, S. Alves Júnior, Use of luminescent gunshot residues markers in forensic context, *Forensic Sci. Int.* 244 (2014) 276–284, doi:<http://dx.doi.org/10.1016/j.forsciint.2014.09.001>.

[19] M.A.M. Lucena, M.F.L. Oliveira, A.M. Arouca, M. Talhavini, E.A. Ferreira, S.A. Júnior, F.H. Veiga-Souza, I.T. Weber, Application of the MOF [Eu(BTC)] as a luminescent marker for GSR: a synthesis, characterization and toxicity study, *Appl. Mater. Interfaces* 9 (5) (2017) 4684–4691, doi:<http://dx.doi.org/10.1021/acsami.6b13474>.

[20] M. López-López, C. Álvarez, J. Pisonero, C. García-Ruiz, N. Bordel, Visualization of gunshot residue patterns by LIBS imaging, *Forensic Sci. Int.* 272 (2017) 124–131, doi:<http://dx.doi.org/10.1016/j.forsciint.2017.02.012>.

[21] M. López-López, C. García-Ruiz, Recent non-chemical approaches to estimate the shooting distance, *Forensic Sci. Int.* 239 (2014) 79–85, doi:<http://dx.doi.org/10.1016/j.forsciint.2014.03.023>. M.A.M. Lucena et al. / *Forensic Science International* 280 (2017) 95–102